

OK to enter - KAC
2/5/07

Serial No. 09/938,623

Atty. Dkt. No. 035879-0125

Amendments to the Claims:

This listing of claims will replace all prior versions, and listings, of claims in the application:

Listing of Claims:

1. (Previously presented) A procytotoxin comprising a cytotoxic peptide bound to an inactivator via a peptide bond, wherein said cytotoxic peptide is a pore-forming cytolytic peptide that comprises an amphipathic alpha-helical structure, and wherein said peptide bond is susceptible to cleavage by a targeting specific protease.
2. (Original) The procytotoxin of claim 1, wherein said inactivator is selected from the group consisting of a microbead, an amino acid, a peptide, phage and a phage filament.
3. (Original) The procytotoxin of claim 1, wherein said inactivator is added to the C-terminus of said cytotoxic peptide.
4. (Original) The procytotoxin of claim 1, wherein said targeting specific protease is a matrix metalloprotease.
5. (Original) The procytotoxin of claim 1, wherein said targeting specific protease is PSA.
6. (Original) The procytotoxin of claim 1, wherein said targeting specific protease is PSMA.
7. (Original) The procytotoxin of claim 6, further comprising at least one lysine residue bound via a peptide bond to at least one amino acid via the ϵ -amino group of said lysine residue.
8. (Original) The procytotoxin of claim 1, further comprising a targeting molecule.

9. (Original) The procytotoxin of claim 8, wherein said targeting molecule is selected from the group consisting of a molecule that targets the neo-vasculature and an antibody.

10. (Original) The procytotoxin of claim 9, wherein said targeting molecule is an RGD targeting sequence.

11. (Original) The procytotoxin of claim 9, wherein said targeting molecule is a neo-vascular targeting sequence of an anti-fibronectin ED-B antibody.

12. (Cancelled)

13. (Previously presented) The procytotoxin of claim 1, wherein said cytolytic peptide is selected from the group consisting of Ae I, cytolsin of sea anemone, aerolysin, amatoxin, amoebapore, amoebapore homolog from *Entamoeba dispar*, brevinin-1E, brevinin-2E, barbatolysin, cytolsin of *Enterococcus faecalis*, delta hemolysin, diphtheria toxin, El Tor cytolsin of *Vibrio cholerae*, equinatoxin, enterotoxin of *Aeromonas hydrophila*, esculentin, granulysin, haemolysin of *Vibrio parahaemolyticus*, intermediolysin of *Streptococcus intermedius*, the lentivirus lytic peptide, leukotoxin of *Actinobacillus actinomycetemcomitans*, magainin, melittin, membrane-associated lymphotoxin, Met-enkephalin, neokyotorphin, neokyotorphin fragment 1, neokyotorphin fragment 2, neokyotorphin fragment 3, neokyotorphin fragment 4, NK-lysin, paradaxin, perforin, perfringolysin O, theta-toxin, of *Clostridium perfringens*, phalloolysin, phallotoxin, streptolysin, and D,L- α -amino acid cyclic peptides.

14. (Previously presented) The procytotoxin of claim 1, wherein said cytolytic peptide is melittin.

15. (Previously presented) The procytotoxin of claim 14, wherein said cytolytic peptide comprises the following structure: Gly-Ile-Gly-Ala-Val-Leu-Lys-Val-Leu-Thr-Thr-Gly-Leu-Pro-Ala-Leu-Ile-Ser-Trp-Ile-Lys-Arg-Lys-Arg-Gln-[Gln-Gly-Ala-Ile-Gly-Gln-Pro] (residues 1-32 of SEQ ID NOS 1 or 2).

16. (Original) The procytotoxin of claim 15, further comprising a targeting molecule.
17. (Original) A pharmaceutical composition, comprising one or more procytotoxins of claim 15 and a pharmaceutically suitable carrier or excipient.
18. (Previously presented) The procytotoxin of claim 14, wherein said cytolytic peptide comprises the following structure: Gly-Ile-Gly-Ala-Val-Leu-Lys-Val-Leu-Thr-Thr-Gly-Leu-Pro-Ala-Leu-Ile-Ser-Trp-Ile-Lys-Arg-Lys-Arg-Gln-[Gln- Ser-Ser-Phe(or Tyr)-Tyr-Ser-Gly(or Ser)] (residues 1-32 of SEQ ID NOS 3 or 4).
19. (Original) The procytotoxin of claim 18, further comprising a targeting molecule.
20. (Original) A pharmaceutical composition, comprising one or more procytotoxins of claim 18 and a pharmaceutically suitable carrier or excipient.
21. (Original) A pharmaceutical composition, comprising one or more procytotoxins of claim 1 and a pharmaceutically suitable carrier or excipient.
22. (Previously presented) A method for selectively destroying a target cell, comprising contacting the target cell with a procytotoxin, which comprises a cytotoxic peptide bound via a peptide bond to an inactivator, wherein said cytotoxic peptide is a pore-forming cytolytic peptide that comprises an amphipathic alpha-helical structure, and wherein said peptide bond is susceptible to cleavage by a targeting specific protease.
23. (Original) The method of claim 22, wherein said inactivator is selected from the group consisting of a microbead, an amino acid, a peptide, phage and a phage filament.
24. (Original) The method of claim 22, wherein said inactivator is added to the C-terminus of said cytotoxic peptide.
25. (Original) The method of claim 22, wherein said targeting specific protease is a matrix metalloprotease.

26. (Original) The method of claim 22, wherein said targeting specific protease is a PSA.

27. (Original) The method of claim 22, wherein said targeting specific protease is a PMSA.

28. (Original) The method of claim 27, wherein said procytotoxin further comprises at least one lysine residue bound via a peptide bond to at least one amino acid via the ϵ -amino group of said lysine residue.

29. (Original) The method of claim 22, wherein said procytotoxin further comprises a targeting molecule.

30. (Original) The method of claim 29, wherein said targeting molecule is selected from the group consisting of a molecule that targets the neo-vasculature and an antibody.

31. (Original) The method of claim 30, wherein said targeting molecule is an RGD targeting sequence.

32. (Original) The method of claim 30, wherein said targeting molecule is a neo-vascular targeting sequence of an anti-fibronectin ED-B antibody.

33. (Cancelled)

34. (Previously presented) The method of claim 22, wherein said cytolytic peptide is selected from the group consisting of Ae I, cytolsin of sea anemone, aerolysin, amatoxin, amoebapore, amoebapore homolog from *Entamoeba dispar*, brevinin-1E, brevinin-2E, barbatolysin, cytolsin of *Enterococcus faecalis*, delta hemolysin, diphtheria toxin, El Tor cytolsin of *Vibrio cholerae*, equinatoxin, enterotoxin of *Aeromonas hydrophila*, esculentin, granulysin, haemolysin of *Vibrio parahaemolyticus*, intermedilysin of *Streptococcus intermedius*, the lentivirus lytic peptide, leukotoxin of *Actinobacillus actinomycetemcomitans*, magainin, melittin, membrane-associated lymphotoxin, Met-enkephalin, neokyotorphin, neokyotorphin fragment 1, neokyotorphin fragment 2, neokyotorphin fragment 3, neokyotorphin fragment 4, NK-lysin, paradaxin, perforin,

perfringolysin O, theta-toxin, of *Clostridium perfringens*, phallolysin, phalloxin, streptolysin, and D,L- α -amino acid cyclic peptides.

35. (Previously presented) The method of claim 22, wherein said cytolytic is melittin.

36. (Previously presented) The method of claim 35, wherein said cytolytic peptide comprises the following structure: Gly-Ile-Gly-Ala-Val-Leu-Lys-Val-Leu-Thr-Thr-Gly-Leu-Pro-Ala-Leu-Ile-Ser-Trp-Ile-Lys-Arg-Lys-Arg-Gln-[Gln-Gly-Ala-Ile-Gly-Gln-Pro] (residues 1-32 of SEQ ID NOS 1 or 2).

37. (Original) The method of claim 36, further comprising a targeting molecule.

38. (Previously presented) The method of claim 35, wherein said cytolytic peptide comprises the following structure: Gly-Ile-Gly-Ala-Val-Leu-Lys-Val-Leu-Thr-Thr-Gly-Leu-Pro-Ala-Leu-Ile-Ser-Trp-Ile-Lys-Arg-Lys-Arg-Gln-[Gln-Ser-Ser-Phe(or Tyr)-Tyr-Ser-Gly(or Ser)] (residues 1-32 of SEQ ID NOS 3 or 4).

39. (Original) The method of claim 38, further comprising a targeting molecule.

40. (Currently Amended) A method of making a procytotoxin, comprising modifying a cytotoxic peptide to include an inactivator wherein said cytotoxic peptide is a pore-forming cytolytic peptide that comprises an amphipathic alpha-helical structure.

41. (Original) The method of claim 40, wherein said inactivator is selected from the group consisting of a microbead, an amino acid, a peptide, phage and a phage filament.

42. (Original) The method of claim 40, wherein said inactivator is added to the C-terminus of said cytotoxic peptide.

43. (Original) The method of claim 40, wherein said targeting specific protease is a matrix metalloprotease.

44. (Original) The method of claim 40, wherein said targeting specific protease is a PSA.

45. (Original) The method of claim 40, wherein said targeting specific protease is a PMSA.

46. (Original) The method of claim 45, wherein said procytotoxin further comprises at least one lysine residue bound via a peptide bond to at least one amino acid via the ϵ -amino group of said lysine residue.

47. (Original) The method of claim 40, wherein said procytotoxin further comprises a targeting molecule.

48. (Original) The method of claim 47, wherein said targeting molecule is selected from the group consisting of a molecule that targets the neo-vasculature and an antibody.

49. (Original) The method of claim 48, wherein said targeting molecule is an RGD targeting sequence.

50. (Original) The method of claim 48, wherein said targeting molecule is a neo-vascular targeting sequence of an anti-fibronectin ED-B antibody.

51. (Cancelled)

52. (Previously presented) The method of claim 40, wherein said cytolytic peptide is selected from the group consisting of Ae I, cytolysin of sea anemone, aerolysin, amatoxin, amoebapore, amoebapore homolog from *Entamoeba dispar*, brevinin-1E, brevinin-2E, barbatolysin, cytolysin of *Enterococcus faecalis*, delta hemolysin, diphtheria toxin, El Tor cytolysin of *Vibrio cholerae*, equinatoxin, enterotoxin of *Aeromonas hydrophila*, esculentin, granulysin, haemolysin of *Vibrio parahaemolyticus*, intermedilysin of *Streptococcus intermedius*, the lentivirus lytic peptide, leukotoxin of *Actinobacillus actinomycetemcomitans*, magainin, melittin, membrane-associated lymphotoxin, Met-enkephalin, neokyotorphin, neokyotorphin fragment 1, neokyotorphin fragment 2, neokyotorphin fragment 3, neokyotorphin fragment 4, NK-lysin, paradaxin, perforin, perfringolysin O, theta-toxin, of *Clostridium perfringens*, phallolysin, phalloxin, streptolysin, and D,L- α -amino acid cyclic peptides.

53. (Previously presented) The method of claim 40, wherein said cytolytic peptide is melittin.

54. (Previously presented) A method of making a procytotoxin, comprising modifying a cytotoxic peptide to include an inactivator, wherein said cytotoxic peptide is a pore-forming cytolytic peptide and wherein said pore-forming cytolytic peptide comprises the following structure: Gly-Ile-Gly-Ala-Val-Leu-Lys-Val-Leu-Thr-Thr-Gly-Leu-Pro-Ala-Leu-Ile-Ser-Trp-Ile-Lys-Arg-Lys-Arg-Gln-[Gln-Gly-Ala-Ile-Gly-Gln-Pro] (residues 1-32 of SEQ ID NOS 1 or 2).

55. (Original) The method of claim 54, further comprising adding a targeting molecule to said procytotoxin.

56. (Original) The method of claim 55, further comprising adding a targeting molecule to said procytotoxin.

57. (Previously presented) The method of claim 53, wherein said cytolytic peptide comprises the following structure: Gly-Ile-Gly-Ala-Val-Leu-Lys-Val-Leu-Thr-Thr-Gly-Leu-Pro-Ala-Leu-Ile-Ser-Trp-Ile-Lys-Arg-Lys-Arg-Gln-[Gln- Ser-Ser-Phe(or Tyr)-Tyr-Ser-Gly(or Ser)] (residues 1-32 of SEQ ID NOS 3 or 4).

58. (Original) The method of claim 40, wherein said target cell is a cancer cell.

59. (Original) The method of claim 58 wherein said cancer cell is selected from the group consisting of prostate, ovarian, breast, skin, lung and pancreas.

60. (Original) A method of treating a cancer patient, comprising administering a therapeutically effective amount of the pharmaceutical composition of claim 21.